Cultivation of *Pleurotus ostreatus* (Oyster Mushroom) Using Sawdust Supplemented with Waste Human Hair

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Abstract: Waste human hair (WHH) is a part of municipal solid waste generated from salons that may lead to clogging of drainage pipes ensued with flooding. *Pleurotus ostreatus* (mushroom) is capable of utilizing several organic substrate types due to its enzyme secretions. This study determined the potential of WHH in cultivating edible mushrooms *P. ostreatus* (Oyster mushroom).The WHH from males was collected from two (2) barbing salons in Benin, Edo State, Nigeria. Microbial analyses of WHH were based on standard methods, while pulverised WHH samples (pasteurised and unpasteurised) were mixed with sawdust in varying concentrations (5, 10, 15 and 20% WHH) and used to monitor the growth of *P. ostreatus* for 100 days. Growth of *P. ostreatus* in sawdust and rice bran was used as control. The total bacterial and fungal counts obtained from WHH samples were $4.50 \pm 1.00 \times 10^2$ cfu/g and 6.10 ± 0.05 x10 sfu/g respectively. Among identified bacteria and fungi were *Bacillus* sp, *Citrobacter* sp. *Penicillium* sp. and *Alternaria* sp. Mycelium complete run observed in the pasteurised samples gave rise to mushrooms with the exception of the 20 % WHH sample. However, the control had the largest total yield (355 g) and biological efficiency (51%) this was followed by the 10% WHH group at 313 g (45%) and the 5% WHH group at 259g (37%). The least values were observed in the 15% WHH group at 87 g (12%) while mushroom growth was not detected in 20 % WHH group. The potential for recycling WHH provides an avenue for the promotion of the circular economy in Nigeria.
Key word: Waste human hair*; Pleurotus ostreatus*, bacteria, fungi, mycelium

INTRODUCTION

The development of cities and generation of wastes are interrelated. The use of dumpsites and landfills remain the popular methods for dealing with wastes across Nigerian city centers (Okwesili *et al*., 2016). Urbanization which is caused mainly by the increase in the number of inhabitants and the level of consumption of such individuals, lead to generation of ever-growing amounts of wastes (Adejobi *et al*., 2012). It is estimated that by 2050, 6 billion people will be living in urban centres which is equal to the world population as at the year 2000. It is forecasted that the volume of municipal solid waste (MSW) will double from 1.3 billion tons per year as at 2012 to 2.2 billion tons per year by 2025 (Oteng-Ababio *et al*., 2018).

An often-overlooked part of municipal solid waste is hair waste generated from barbing salons. Hair fibre is the non-living part of hair that is trimmed from our scalps (Pergament, 2018). Keratin is its major constituent and it makes up about 65%-95% of the hair weight. Other constituents include water, lipids, pigment and trace elements (Cruz *et al*., 2016). In rural areas, the hair fibre is often thrown away in nature where it slowly decomposes over several years, eventually returning the constituent elements such as carbon, nitrogen, sulphur and minerals to their respective natural cycles (Cruz *et al*., 2016). In urban areas however, hair accumulates in barbershops and are then thrown away at dumpsites where they accumulate or spill into the drainage systems causing clogging of drainage pipes (Gupta *et al*., 2014). Due to its recalcitrant nature, hair fibres stay in dumpsites occupying large volumes of space.

Overtime, leachate from these dumps increase the nitrogen concentration in nearby water bodies, causing eutrophication (Adejobi *et al*., 2012). Further, burning of human hair fibre leads to the release of foul odour and toxic gases such as ammonia, carbonyl and hydrogen sulphides (Dimuna, 2004; Gupta *et al*., 2014). Again, open dumps of hair generate hair dust, which causes discomfort to individuals living close to them and if inhaled in large amounts, can

result in severe respiratory problems like asthma, pulmonary fibrosis, and lung cancer (Dimuna, 2004). The best way to address such problems is to develop systems which utilize the waste material as a resource and in addition contribute to economic growth (Gupta, 2014). Bacteria profiling of hair was demonstrated to be a useful forensic tool to augment existing forensic techniques (Tridico *et al*., 2014). The bacterial community structure of the human hair shaft has been profiled by thermal gradient gel electrophoresis (TGGE), which revealed approximately 20 bacterial species including *Pseudomonas* sp. (Tridico *et al*., 2014). Again, a metagenomics analysis of bacteria on human scalp hair revealed high diversity with 4,838 core bacteria and 1,220 transient bacteria (Kerk *et al*., 2018). Bacteria such as *Staphylococcus aureus* and *Staphylococcus epidermidis* as well as *Escherichia coli* have been identified on human skin and scalp (Brooke *et al*., 2009; Kerk *et al*., 2018). *Aspergillus*, *Paecilomyces* and *Penicillium* were among fungal isolates from human hair samples obtained from different governorates in Egypt (Gherbawy *et al*., 2006).

Pleurotus ostreatus (mushrooms) are fleshy, spore-bearing reproductive structures of fungi grown on organic substrates (Besufekad *et al*., 2020). The mushroom plays an important role as food for humans, due to their nutritional and medicinal properties. Oyster mushrooms have been reported to contain protein, fat, sugars, mycocellulose, and minerals (Mahadevan and Shanmugasundaram, 2018). Since Oyster mushrooms can be grown on various substrates, including paddy straw and maize stalks/cobs, hence could play an important role in managing recalcitrant organic wastes such as waste human hair (Carrasco *et al*., 2018). Therefore, waste human hair fibre known to contain amino acids and minerals can be used as a nitrogen supplement to help boost mushroom growth. The study investigated the cultivation of *Pleurotus ostreatus* (oyster mushroom) using sawdust supplemented with waste human hair.

MATERIALS AND METHODS

Sample Collection: Waste human hair (WHH) from male scalps were collected from 2 hair salons situated in Benin City, Edo State, Nigeria. A large plastic sterile bin was used for hair sample collection. *Pleurotus ostreatus* mycelium was obtained from Mycofarms and Allied Synergy Limited, Benin City, Edo State, Nigeria.

Preparation of Culture Media for Microbial Analysis: Freshly collected hair samples were picked with flamed tongs from the special bins and placed in 4 sterile sample bottles and sent to the Laboratory for microbial analysis. Media used were nutrient agar (NA) and potato dextrose agar (PDA) for bacterial and fungal isolation respectively. Media were prepared according to manufacturer's instructions (Shruti *et al*., 2022).

Bacterial Isolation and Identification of Waste Human Hair: One (1) g of WHH was weighed and placed in 9 ml sterile water and allowed to stand for 30 minutes. The suspensions were diluted serially and an aliquot (0.1 ml) was then transferred aseptically to sterile Petri plates. Nutrient agar (sterile) was poured in aseptically and plates were incubated at 37°C for 24 - 48 h. After successful growth of bacteria, the colonies were counted with a colony counter and the results per dilution count were recorded in colony forming units (cfu) per gram. Pure cultures were identified and characterized on the basis of their cultural, morphological and biochemical characteristics (Aneja, 2008).

***Fungal Isolation and Identification of Waste Human Hair*:** One gram of hair sample was diluted serially using sterile distilled water. Subsequently, an aliquot of 0.1 ml was transferred aseptically to sterile Petri plates and sterile PDA was poured in aseptically and incubated at 28°C for 72 h. After successful growth of fungi, the number of spore-forming unit per gram was calculated. Fungal isolates identification was based on morphology and microscopy (Aneja, 2008).

Waste human hair sample preparation: The WHH samples were washed and rinsed thoroughly with hexane and methanol (1:1) and subsequently with 70% (v/v) ethanol to remove impurities. The samples were then air-dried for 72 h and 4 kg of waste human hair was then sent to a mill for pulverisation (Singh *et al*., 2019). The pulverised WHH was kept in sterile universal bottles and stored for further use.

Spawn Production of Pleurotus ostreatus: Guinea corn was cleaned manually to remove inert matter, stubble, stones and debris. Two (2) kg of the cleaned grains were soaked in 0.5% $CuSO_4$ for 10 mins, thoroughly washed twice with 10 l of tap water and soaked in 6 l of tap water for 2 h. Thereafter, the grains were drained and the following additives added: rice bran at the rate of 10% and chalk ($CaCO_3$) at the rate of 2% on dry weight basis of the grains. The additives were thoroughly and evenly mixed with the grains. The grain medium was filled halfway into 500 ml glass bottles and a stopper of cotton wool and foil paper/paper towel was used to plug the mouth of the bottles. These were firmly tied with rubber bands around the bottle necks. The bottles (6) containing the grains and additives were autoclaved at 121°C at 15 psi for 30 mins and allowed to cool for 24 h. The bottles were inoculated with *P. ostreatus* mycelium sustained on potato dextrose agar (PDA) and allowed to grow at room temperature with low light for 10 days. Grain spawn was available after 10 (Ten) days of mycelium run (Tesfay *et al*., 2020).

Waste Human Hair Supplementation with Sawdust: The pulverised WHH was mixed with sawdust in 5 different ratios (5, 10, 15 and 20%) both in pasteurised and unpasteurised treatments. Calcium hydroxide (CaOH) was added in varying proportions to all groups containing WHH to serve as a mild protein denaturing agent. The control group had 1520 g of sawdust and 380 g of rice bran, 5% WHH group had 1805 g of sawdust and 95 g of WHH, the 10% WHH group had 1710 g of sawdust and 190 g of WHH while the 15% WHH group had 1615 g of sawdust and 285 g of WHH and finally, 1520 g of sawdust and 380 g of WHH was used for 20% WHH. The different treatment groups were mixed with water for 24 h to moisten it thoroughly. The setup were bagged accordingly and divided into 2 groups. One group was pasteurized at 121°C and 15 psi, for 12 h and after cooling for another 12 h was inoculated with oyster mushroom grain spawn. The other group which was the unpasteurised was also inoculated with oyster mushroom grain spawn after bagging. All treatments were sealed and mycelial growth monitored in a room with low light at room temperature. A meter rule was used to measure mycelium run in the bags. After 40 days of growth, the bags were transferred to the cropping room with limited light and average temperature of 25 °C for 60 days making a total of 100 days between acclimatization and harvest (Tesfay *et al*., 2020). Holes were then cut into the bags to allow for aeration of the mycelium. The growing bags of mycelia were watered twice a day (morning and afternoon) using a spray bottle to maintain water activity of the substrates in the bags and humidity of the cropping room (Tesfay *et al*., 2020).

Mushroom Harvesting and Yield Quantification: The time required from acclimatization to completion of mycelium run were recorded (days) for both treatments (Ashraf *et al*., 2013). However, mushrooms were subsequently harvested in the first (first time a bag yields mushroom) and second (second time a bag yields mushroom) flushes (harvests) from the pasteurized group (Tesfay *et al*., 2020). The total weight of all the fruiting bodies harvested from the two flushes/harvests were measured as total yield of mushroom. The yield parameters were recorded with respect to time (days) taken for completion of mycelium run, time taken for maturity of fruit bodies, number of flushes, and yield of flushes on the treatment substrates (total weight of all the fruiting bodies harvested from the two flushes were measured and considered as total yield of mushroom). The average biological

efficiency (BE) of harvested mushrooms was calculated using the formular below:

$$BE = \frac{\text{Weight of fresh mushroom harvested per bag}}{\text{Weight of dry substrate per bag before inoculation}} \times 100$$

Statistical Analysis*:* Most experiments were performed in replicates and values were reported as means ± standard deviations. Differences in growth were analysed using IBM SPSS statistics package 20 (SPSS Inc. and IBM Company, Chicago, USA) where P < 0.05 implies significant difference between values obtained (Ogbeibu, 2015).

RESULTS

Waste human hair (WHH) collected from 2 hair salons were analysed for bacteria and fungi as well as their impact on the growth of *P. ostreatus.* Table 1 showed that the bacterial counts (4.50±1.00 x10^2 cfu/g) was significantly higher than the fungal counts (6.10±0.10 x10 sfu), while Supplementary Tables 1 and 2 revealed the identity of isolated bacteria and fungi, respectively. Mycelial growth was monitored in different mixtures of sawdust and WHH (5, 10, 15 and 20 %) in both unpasteurized and pasteurized groups for 40 days. Generally, in unpasteurized groups, mycelial run peaked on day 10 in treatments 5%, 15% and 20%, while this was on day 20 in the control and 10% group. By day 40, mycelial growth had dropped significantly (P < 0.05) in all the treatments (Table 2). Conversely, mycelial run was completed on day 30 in the control and 5% groups in pasteurized groups. By day 40, the 10% group had completed mycelial run in their bags however, 15% and 20% groups did not complete mycelium run during the 40 days (Table 2).

Table 1: Total microbial counts of waste human hair collected from barbing salons

	Male Barbing Salon 1		Male Barbing Salon 2		Total Microbial Count
	Sample 1	Sample 2	Sample 1	Sample 2	
Bacteria (x10^2 cfu/g)	3.84	3.46	5.22	5.48	4.50±1.00
Fungi (x10 sfu/g)	8.10	5.40	6.50	6.50	6.10±0.10

Table 2: Mycelium run in unpasteurized and pasteurized substrate (mm)

		Days				
		1	10	20	30	40
Unpasteurised	Control	13.42±1.23^{a}	32.75±1.12^{a}	33.55±1.00^{a,b}	8.89±0.67^{a}	0.02±0.04^{a}
Trts (%)	5	10.7±1.11^{b}	21.77±1.02^{b}	21.61±0.93^{c}	5.22±0.27^{b}	0.06±0.08^{a}
	10	12.26±0.71^{a,b}	32.54±0.72^{a}	32.64±0.76^{a}	8.114±0.59^{a}	2.34±0.79^{b}
	15	12.05±0.75^{a,b}	31.8±1.03^{a}	31.76±1.02^{a}	5.80±0.72^{b}	2.26±0.71^{b}
	20	13.4±0.93^{a}	36.4±1.85^{c}	35.74±1.97^{b}	5.21±0.53^{b}	1.86±0.63^{b}
Pasteurised	Control	22.80±0.66^{a}	86.80±5.42^{a}	193.20±3.21^{a}	200.85±0.20^{a}*	200.85±0.20^{a}*
Trts (%)	5	27.20±1.19^{c}	91.90±2.89^{a}	204.85±2.94^{a}	211.15±0.86^{a}*	211.15±0.86^{a}*
	10	25.44±0.94^{b,c}	58.95±3.64^{b}	140.65±4.07^{b}	178.85±8.31^{b}	200.95±11.74^{a}*
	15	23.64±2.01^{a,b}	42.30±8.50^{c}	82.20±15.06^{c}	107.35±16.06^{c}	121.40±19.33^{b}
	20	24.92±0.95^{a,b,c}	34.35±4.91^{c}	45.40±8.30^{d}	71.00±4.00^{d}	74.50±3.50^{c}

Key: *Mycelium run measurements (mm) were conducted in triplicates, and the numbers represent the average ± SD. Different superscripts within columns indicate significant mean values of triplicate samples that are significantly different (P < 0.05) for unpasteurized and pasteurized substrates (mm).

Plate 1 shows mycelial biomass growing downwards in both treatments at day 3. However, on day 18, the mycelium growth in the unpasteurised group appears to have stalled which was in contrast to continuous mycelium growth observed in the pasteurized group (Plate 1). The control group and the 5% WHH group exhibited quick mycelium growth, while 10, 15 and 20% WHH groups also showed mycelium growth albeit at slower rate in the pasteurized group. On day 30, pasteurized group mycelial run was completed in the control and 5% WHH groups. The 10%

WHH group completed mycelium run on day 40. The 15% and 20% hair group never completed mycelium run. However, browning was observed to occur in the control and 5% group of the unpasteurized, and stunted mycelial growth was observed in all groups of the unpasteurised substrates. (Plate 1). The weight of harvested mushrooms (first flush/harvest) from 42 to 91 d across pasteurized group is shown in Table 3 and the dry weight counterpart is shown in Table 3. The control group had the largest mean weight followed by the 10% WHH group (Table 3). Plate 2 shows that the 5% WHH bag sprouted mushrooms before the control group. Subsequently, the weight of the mushrooms harvested from the second flush (harvest) from 72 to 100 d in pasteurized group is shown in Table 4. Here, the control group had the mean weightiest mushroom which was closely followed by the 10% WHH group. The mean dry weight of these mushrooms is shown in Table 4.

Plate 1: Mycelium run across the unpasteurised treatment and pasteurised treatment at 3, 18 and 30 days. A - Unpasteurised treatment; B - Pasteurised treatment

Table 3: Weight (g) and dry weight (g) of mushrooms harvested from the pasteurised substrates 1st Flush (42-91 days)

	Treatments (%)				
Weight (g)	Control	5	10	15	20
Bag Mean	186.67±32[a]	136.67±19[a, b]	148.67±26[a, b]	88.67±48[b]	ND
Day of harvest (mean)	62 days	47 days	64 days	74 days	ND
Dry weight (g)					
Bag Mean	18.00±1.00[a]	11.67±1.53[b]	11.33±1.16[b]	8.67±3.79[b]	ND
Day of harvest (mean)	62 days	47 days	64 days	74 days	ND

Key: *Mushrooms were harvested in triplicates, and the numbers represent the average ± SD. Different superscripts across the row indicate mean values of triplicate samples that are significantly different ($P < 0.05$) of weight and dry weight (g) of mushrooms. ND - Not Detected

Table 4: Weight (g) and dry weight (g) of mushrooms harvested from the pasteurised substrates 2nd Flush (72-100 days)

	Treatments (%)				
Weight (g)	Control	5	10	15	20
Bag Mean	147.33±16.16[a]	94.00±6.25[b]	136.33±19.14[a]	39.00±18.52[c]	ND
Day of harvest (mean)	90 days	93 days	90 days	96 days	ND
Dry weight (g)					
Bag Mean	11.33±3.06[a]	10.67±0.58[a]	12.00±1.00[a]	5.00±2.65[b]	ND
Day of harvest (mean)	90 days	93 days	90 days	96 days	ND

Key: *Mushrooms were harvested in triplicates, and the numbers represent the average ± SD. Different superscripts across the row indicate mean values of triplicate samples that are significantly different ($P < 0.05$) of weight and dry weight (g) of mushrooms. ND - Not Detected

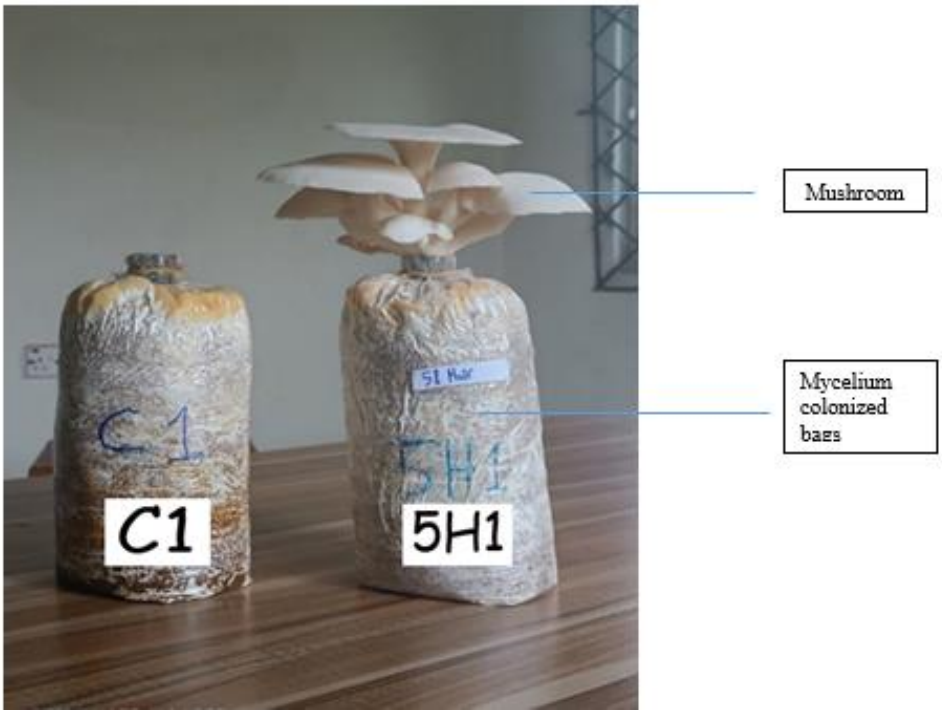

Plate 2: First flush from 5% hair group (5H 1) sprouted earlier on Day 47 relative to Control (C1)

Table 5: Total yield (g) and biological efficiency (%) of oyster mushroom across pasteurized group

		Treatments (%)			
Weight (g)	Control	5	10	15	20
Bag 1 (g)	329g (47%)	209g (30%)	276g (39%)	204g (29%)	ND
Bag 2 (g)	324g (46%)	224g (32%)	313g (45%)	87g (12%)	ND
Bag 3 (g)	355g (51%)	259g (37%)	266g (38%)	92g (13%)	ND

Key: ND - Not Detected

Plate 3: Mushroom mycelial thread comparison in mushrooms from A (control), group B (5% WHH group) *A - Mycelial Thread + Rice bran + Sawdust; B - Mycelial Tread + Waste Human Hair (WHH) + Sawdust

The harvested mushrooms possessed a network of mycelial threads that could be observed in Plate 3 which showed waste human hair (WHH) in the mycelial thread matrix. The mushrooms grown on rice bran + sawdust (control) showed brownish composites while that grown on human hair + sawdust (5% WHH group) showed presence of black string like substances (hair strands) in the mycelium. This goes to show that mycelium absorbed the WHH and locked it into its matrix.

DISCUSSION

Waste human hair (WHH) from male salons were analysed for bacteria and fungi as well as their impact on the growth of *P. ostreatus*. The WHH was found to contain two (2) Gram positive (*Staphylococcus aureus* and *Bacillus* sp.) and two (2) Gram negative bacteria (*Klebsiella* sp. and *Citrobacter* sp). Human hair follicle contains plethora of bacteria, fungi and viruses that changes sometimes in disease conditions, and include both Gram +ve and Gram -ve bacteria found in scalp hair and skin (Lousada *et al*., 2021). Furthermore, geographic location as well as lifestyle can influence the microbial community present on the hair shaft and follicle. As the human scalp hair interacts with environment consistently, the composition and type of bacteria found in the hair will also vary (Brinkac *et al*., 2018). Bacteria has been reported to respond differently to human hair shaft for example, *P. aeruginosa* and *E. coli* adhered to and colonized human hair shafts without significantly affecting the hair shaft's surface morphology, while *E. coli* inhabited the edges of the cuticle scales (Kerk *et al*., 2018). Four fungal isolates were also obtained from WHH which included *Penicillium* sp., *Rhizopus* sp., *Aspergillus* sp. and *Alternaria* sp. The moist nature of the human scalp, makes it an ideal area for certain fungal populations to grow (Skowron *et al*., 2021) *Aspergillus*, *Penicillium* and *Paecilomyces* were among fungal isolates from human hair samples obtained from different governorates in Egypt (Gherbawy *et al*., 2006). However, in mushroom farming, these microorganisms may compete with *P. ostreatus* (oyster mushroom) for nutrition and can hinder the growth of the mushroom, leading to loss of profits (Jang *et al*., 2019).

After bacterial and fungal isolation from WHH, mycelial growth of *P. ostreatus* was monitored in different mixtures of sawdust and WHH (5, 10, 15 and 20 %) in both pasteurized and unpasteurized groups for 40 days. The unpasteurized group (control, 5, 10, 15 and 20% treatments) did not complete mycelium run and by day 40, mycelium run had drastically reduced and a brownish discolouration was observed in the bags. The unpasteurized substrates did not yield any mushrooms due to incomplete mycelium run. This could be as a result of competing microbes that negatively impacted the growth of Oyster mushrooms in the unpasteurized treatments. This is in agreement with work of Lim *et al*. (2008) where *Bacillus* sp isolated from diseased *Pleurotus eryngii* (King Oyster) showed inhibitory activity on the mycelium growth of King Oyster. Similarly, Jang *et al*. (2019) observed that the presence of *Aspergillus* sp. and *Penicillium* sp. in mushroom farms caused devastation to mushroom harvest. Conversely, in the pasteurized substrates, mycelium run was completed in the control group and 5% hair group first which was subsequently followed by the 10% hair group however, 15% and 20% hair group never completed mycelium run. This could be as a result of the fact that no microorganisms were competing with *P. ostreatus* for growth as a result of the pasteurization process.

After mycelium run for 40 days, mushrooms began to sprout between 41 to 100 days and were harvested in 2 flushes in the pasteurized group. Different treatment bags (Control, 5, 10 and 15%) sprouted mushrooms randomly within 60 days (41-100 days). In the first flush (harvest), the 5% hair group had the earliest mushroom sprout. This could be attributed to the availability of

waste human hair in adequate amount to stimulate mushroom growth. Again, the increased surface area of milled human hair waste may have made it more accessible to the mushroom for their immediate growth. The 10% hair group had the second highest average mushroom weight that was only surpassed by the control group. This is probably due to the introduction of a new substrate (human hair) at higher concentration to *P. ostreatus* which requires more time for adaptation. However, in the second flush, the average weight of the mushrooms harvested from the 10% hair group was not significantly different from that harvested from the control. This could be as a result of reduced nutrient availability in both treatments due to the first harvest. This is also similar to the work carried out by Tesfay *et al*. (2020) where the second flush of *P. ostreatus* mushrooms growth on waste paper and corn stalk, weighed less than the first flush.

The control group had the highest average biological efficiency at 51%, followed by the 10% and 5% hair group at 45% and 37% respectively. For a first-time substrate, waste human hair was able to compete with rice bran (plant-based nitrogen source) in the control group and supplemented sawdust to yield mushrooms. The WHH been an organic nitrogen source may have caused mushroom yields in the 5%, 10% and 15% hair group, and this has shown that animal-based organic nitrogen source can be exploited for mushroom growth (Cruz *et al*., 2016). This is similar to the findings of Carrasco *et al*. (2018) who reported the use of feather flour as a nitrogen supplement in mushroom farms in Brazil and some parts of Europe. From their research, it was observed that mushroom farmers used feather flour as an alternative nitrogen source to existing supplements which are relatively expensive. Furthermore, Taskin *et al*. (2012) observed that mycelial biomass and extracellular polysaccharides of *Rhodotorula glutinism* increased significantly when supplemented with chicken feather hydrolysate. The authors were able to supplement the yeast specie (*Rhodotorula glutinis*) with a low-cost peptone alternative in the form of chicken feather hydrolysate. Again, it was observed that the fungi, *Isaria farinosa* grew significantly better when supplemented with beef extract as compared to inorganic nitrogen sources (Liu *et al*., 2018). Again, Chioza and Ohga, (2013) observed that *Paecilomyces hepiali* preferred organic sources of nitrogen like peptone and beef extract compared to inorganic sources like urea. The use of varying substrate types regularly for edible fungi cultivation preserves its biodiversity (Stamets, 2000) Therefore, diverse substrate types will encourage the production of various enzymes in the extracellular matrix, allowing for the fungi to thrive on numerous types of substrates. Biodiversity ensures that species can thrive despite changes in their environment. In the case of edible mushrooms, multiple substrate types would allow for the survival of edible mushroom species over several generations as well as preserve their extracellular enzymatic activity. Although, literature on the use of human hair as a substrate for the growth of mushrooms is limited. This study serves as a precursor for subsequent work in the use of organic wastes of animal origin in the creation of feedstock for mushroom growth and development.

CONCLUSION

The presence of competing microbes reduced the growth potential of Oyster mushroom in unpasteurised substrates. The luxuriant growth of *P. ostreatus* (oyster mushroom) mycelium was observed in the pasteurised control, 5, 10 and 15% hair groups. The 5% hair group had the earliest mushroom sprouting, while the control group had the highest yield and biological efficiency which was closely followed by the 10% hair group. The composition of mushroom growth substrate recipes could determine the extent of its growth. Conclusively, the growth of *P. ostreatus* in sawdust supplemented with waste human hair was observed, and this adds to the

variety of mushroom growth recipes currently available across the globe. Therefore, the inclusion of WHH in the cultivation of *P. ostreatus* and by extension other edible mushrooms could help in the management and utilization of WHH, thus alleviate its menace in the environment.

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REFERENCES

Adejobi, O. S., and Olorunnimbe, R. O. (2012). Challenges of waste management and climate change in Nigeria: Lagos State metropolis experience. *African Journal of Science Research*, 7(1), 346-362.

Aneja K.R. (2008). A Textbook of Basic and Applied Microbiology, New Age International Publishers, New Delhi.

Ashraf, J., Asif, A. M., Ahmad, W, Ayyub, M. C. and Shafi, J. (2013). Effect of different substrate supplements on Oyster Mushroom (*Pleurotus* spp.) Production. *Food Science and Technology*, 1(3), 44-51.

Besufekad, Y., Mekonnen, A., Girma, B., Daniel, R., Tassema, G., Melkamu, J., Asefa, M., Fikiru, T. and Denboba, L. (2020). Selection of appropriate substrate for production of oyster mushroom (*Pleurotus ostreatus*). *Journal of Yeast and Fungal Research*, 11(1), 15-25.

Brinkac, L., Clarke, T. H., Singh, H., Greco, C., Gomez, A., Torralba, M. G., Frank B and Nelson, K. E. (2018). Spatial and Environmental Variation of the Human Hair Microbiota. *Scientific Reports*, 8, 9017.

Brooke, J. S., Annand, J. W., Hammer, A., Dembkowski, K., and Shulman, S. T (2009). Investigation of bacterial pathogens on 70 frequently used environmental surfaces in a large urban U.S. University. *Journal of Environmental Health*, 71, 17–22.

Carrasco, J., Zied, D. C., Pardo, J. E., Preston, G. M. and Pardo-Giménez, A. (2018). Supplementation in mushroom crops and its impact on yield and quality. *AMB Express*, 8,146.

Chioza, A. and Ohga S. (2013). Mycelial Growth of *Paecilomyces hepiali* in Various Agar Media and Yield of Fruit Bodies in Rice Based Media. *Advances in Microbiology*, 3, 529-536.

Cruz, F. C., Costa, F., Gomes, A., Andreia, C. G., Matama, T. and Cavaco-Paulo A. (2016). Human Hair and the Impact of Cosmetic Procedures: A Review on Cleansing and Shape-Modulating Cosmetics. *Cosmetics*, 3(3),26 https://doi.org/10.3390/cosmetics3030026.

Dimuna, K.O (2004). The Problem with the Management of Environmental Pollution in Nigeria. *Journal of Applied and Basic Sciences*, 2(2), 5-9.

Gherbawy, Y. A. M. H., Maghraby, T. A., El-Sharony, H. M., Hussein, M. A (2006). Diversity of Keratinophilic Fungi on Human Hairs and Nails at Four Governorates in Upper Egypt. *Mycobiology*, 34(4), 180-184.

Gupta, A (2014). Human Hair Waste and Its Utilization: Gaps and Possibilities. *Journal of Waste Management*, 1-17, Article ID 498018 http://dx.doi.org/10.1155/2014/498018.

Jang, M. J., Park, Y., Kim, J. H. (2019). Properties of disease occurrence by season for cultivation facilities of

oyster mushroom. *Journal of Mushrooms,* 17(3), 93-98.

Kerk, S. K., Lai, H. Y., Sze, S. K., Ng, K. W., Schmidtchen, A. and Adav, S. S (2018). Bacteria Display Differential Growth and Adhesion Characteristics on Human Hair Shafts. *Frontiers in Microbiology*, 9, 21-22.

Lim, Y., Ryu, J. S., Shi, S., Noh, W., Kim, E., Le, Q. V., Lee, H. S., Ro, H. S. (2008) Isolation of bacteria associated with the king Oyster Mushroom, *Pleurotus eryngii*. *PubMed Central*, 36(1), 13-18.

Liu, F., Xiang, M., Guo, Y., Wu, X., Lu, G., Yang, Y., Liu, X., Chen, S., Zhang, G. and Shi W. (2018). Culture conditions and nutrition requirements for the mycelial growth of *Isaria farinosa* (*Hypocreales: Cordycipitaceae*) and the altitude effect on its growth and metabolome. *Scientific Reports*, 23:8(1), 15623.

Lousada, M. B., Lachnit, T., Edelkamp, J., Rouillé, T., Ajdic, D., Uchida, Y., Di Nardo, A., Bosch, T. C. G. and Paus, R. (2021). Exploring the human hair follicle microbiome. *British Journal of Dermatology,* 184 (5), 802–815.

Mahadevan, K. and Shanmugasundaram K. (2018). Comparative effect of different culture media on mycelial growth performance of *Pleurotus sapidus. Journal of Pharmacognosy and Phytochemistry*, **7**(4), 874-878.

Ogbeibu, A. R (2015). Biostatistics: A Practical Approach to Research and Data Handling (2nd ed.). Mindex Publishing Company, Lagos, Nigeria. 165pp.

Okwesili, J., Ndukwe, C. and Nwuzor, C. I. (2016). Urban Solid Waste Management and Environmental Sustainability in Abakaliki Urban, Nigeria. *European Scientific Journal*, 12(23), 155-183.

Oteng-Ababio, M., Annepu, R., Bourtsalas, A., Intharathirat, R. and Charoenkit, S (2018). Urban solid waste management. In Rosenzweig, C., W. Solecki, P. Romero-Lankao, S. Mehrotra, S. Dhakal, and S. Ali Ibrahim (eds.), Climate Change and Cities: Second Assessment Report of the Urban Climate Change Research Network. Cambridge University Press. New York. 553–582.

Pergament, D. (2018). It's Not Just Hair: Historical and Cultural Considerations for an Emerging Technology. *Chicago-Kent Law Review*, 75(1), 1-21.

Shruti, T. M., Ramamoorthy, V. and Vellaikumar, S. (2022). Effect of different culture media, temperature and pH on the mycelial growth of *Pleurotus eryngii* (King Oyster Mushroom). *The Pharma Innovation Journal,* 11(7), 2519-2522.

Singh, T. and Prasad K. (2019). Multi-tasking hydrated ionic liquids as sustainable media for the processing of waste human hair: a biorefinery approach. *Green Chemistry*, 21, 3328–3333.

Skowron, K., Bauza-Kaszewska, J., Kraszewska, Z., Wiktorczyk-Kapischke, N., Grudlewska-Buda, K., Kwiecińska-Piróg, J., Wałecka-Zacharska, E., Radtke, L. and Gospodarek-Komkowska, E. (2021). Human Skin Microbiome: Impact of Intrinsic and Extrinsic Factors on Skin Microbiota. *Microorganisms*, 9(3), 543.

Stamets P. (2000). Growing Gourmet and Medicinal Mushrooms. 3rd edition, Ten Speed Press, California. 87p

Taskin, M., Ozkan, B., Atici, O. and Aydogan, M. N (2012). Utilisation of chicken feather hydrolysate as a novel fermentation substrate for production of exopolysaccharides and mycelial biomass from edible mushroom Morcella esculenta. *International Journal of Food Sciences and Nutrition*, 63(5), 597-602.

Tesfay, T., Godifey, T., Mesfn, R. and Kalayu G. (2020). Evaluation of waste paper for cultivation of oyster mushroom (*Pleurotus ostreatus*) with some added supplementary materials. *AMB Express*, 10, 15.

Tridico, S. R., Murray, D. C., Addison, J., Kirkbride, K. P. and Bunce, M. (2014). Metagenomic analyses of bacteria on human hairs: a qualitative assessment for applications in forensic science. *Investigative Genetics*, 5(16), 1-14.